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Original Article

Serum miR-195: function as a potential diagnostic biomarker in Chinese septic patients

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Abstract: Sepsis is serious systemic inflammatory response syndrome (SIRS), which often leads to severe inflammation and organ dysfunction. Herein, we explored and analyzed the expression level of serum miR-195 in sepsis and its diagnostic value. 54 septic patients and 41 SIRS patients were recruited for sera samples in the present study. qRT-PCR was performed to detect the levels of serum miR-195 in two cohorts of patients. The diagnostic value of serum miR-195 in sepsis was evaluated and analyzed using ROC curve and survival analysis was performed to analyze and evaluate the association between the expression levels of serum miR-195 and prognostic outcomes of septic patients. miR-195 serum expression in septic patients was markedly increased in comparison to that in SIRS patients ($P<0.001$), but there was no significant association between serum miR-195 levels and infection sites of septic patients. The area under the curve (AUC) of serum miR-195 expression levels between sepsis group and control group was 0.776, and its specificity was 70.37% and sensitivity was 75.61%. Serum miR-195 expression was positively correlated with APACHE II scores in septic patients. The septic patients with high miR-195 level had a relatively lower 28 d survival rate, compared with patients with low miR-195 level, but the difference was not statistically significant ($P=0.113$). The expression level of serum miR-195 is elevated in sepsis and serum miR-195 could be a potential diagnostic biomarker for the septic patients.

Keywords: miR-195, serum, sepsis, biomarker, diagnosis

Introduction

Sepsis, as a fatal systemic disease in the intensive care unit (ICU), is commonly triggered through the microbial infection, leading to an aberrant activation of the innate immune system [1]. One clinical characteristic of sepsis is serious systemic inflammatory response syndrome (SIRS), which could impair the normal function of lungs, brain, kidneys and other organs [2-5]. Despite great achievement in diagnostic and therapeutic methods, the mortality rates of sepsis remain at a high level ranging from 30 to 70% worldwide [6]. Recently, procalcitonin (PCT) often serves as a helpful diagnostic biomarker for sepsis, but its accuracy in critically ill patients still remains divergent [7]. Accordingly, it is in urgent necessary for us to further elucidate the underlying mechanisms of pathogenesis and progression of sepsis and provide novel, promising diagnostic and therapeutic strategies for patients with sepsis [8].

MicroRNAs (miRNAs), also known as non-coding, single stranded, small RNA molecules (~22 nt), could regulate expression of downstream genes via directly binding with 3'-untranslated region (3'-UTR) of target messenger RNAs (mRNAs) [9]. During the past decades, miRNAs are considered to regulate more than 50% of protein-coding genes and a number of biological processes since each miRNA participates in regulating hundreds of downstream target genes [10]. In addition, the aberrant expression of miRNAs might cause severe dysfunction of immune system, thus leading to the occurrence and development of autoimmunity diseases, leukaemia and solid tumors [11]. The recent studies revealed that aberrant miRNAs expression was associated with sepsis [12, 13]. Because of stable expression in plasma or serum, a number of circulating miRNAs, including miRNA-143 [14], miR-23a-5p [15], miR-133a [16] are applied as diagnostic and prognostic biomarkers in sepsis. Recently, increas-

Table 1. The demographic characteristics of sepsis group and control group

Characteristics	Sepsis group (n=54)	Control group (n=41)	*P* value
Age (year)	60.4 ± 5.8	61.6 ± 5.7	0.716
Gender			
Male	31 (57.4%)	22 (53.7%)	0.712
Female	23 (42.6%)	19 (46.3%)	
Ethnicity			
Han	48 (88.9%)	32 (78.0%)	0.151
Minority	6 (11.1%)	9 (22.0%)	
Registered residence			
Urban	27 (50.0%)	18 (43.9%)	0.555
Rural	27 (50.0%)	23 (56.1%)	
Years of education			
>9	24 (44.4 %)	15 (36.6%)	0.441
≤9	30 (55.6%)	26 (63.4%)	
Marital status			
Married	44 (80.0%)	34 (82.9%)	0.716
Unmarried	11 (20.0%)	7 (17.1%)	

Table 2. Infection sites of septic patients

Infection site	Septic patients (n=54)
Pulmonary	25 (46.3%)
Abdominal	11 (20.4%)
Urological	4 (7.4%)
Brain-derived	2 (3.7%)
Other	12 (22.2%)

ing evidences indicated that aberrant expression of miR-195 serve a critical role in a variety of diseases. For example, Min *et al.* [17] demonstrated that miR-195 could bind with the 3'-untranslated region (3'UTR) of AQP8 and aggravate inflammation in the pathogenesis of ulcerative colitis (UC).

To our knowledge, this is the first article to explore the association between serum miR-195 and sepsis. In the present study, we aimed to investigate the levels of serum miR-195 in septic patients. The further investigations were performed to evaluate whether serum miR-195 could be regarded as a promising biomarker in sepsis diagnosis.

Materials and methods

Study design

In our study, 54 patients with sepsis were recruited from department of intensive care unit (ICU) at the first affiliated hospital during a period from May 2014 to November 2015. In addition, 41 patients with SIRS were recruited as controls from outpatient service of the first affiliated hospital. In the present study, a sepsis was defined as the presence of SIRS associated with infection. SIRS was diagnosed by the presence of at least two of the following [18]: (a) body temperature >38°C (hyperthermia) or <36°C (hypothermia), (b) P_{co2}<32 mmHg or respiratory rate >20/min, (c) heart rate >90/min, and (d) white blood cells >12,000/mm^3 (leukocytosis) or <4,000/mm^3 (leukopenia). The demographic characteristics of all the participants were recorded in **Table 1**, and the infection sites of septic patients were documented in **Table 2**. There was no remarkable difference in age, gender, ethnicity, registered residence, years of education and marital status between the two groups (all *P*>0.05). Acute Physiology and Chronic Health Evaluation II (APACHE II) score, indicating the severity of condition [19], and in-hospital mortality of the 54 patients through following up for 28 days or until death were also documented.

This study protocol was in accordance with the ethical guidelines of the 1995 Declaration of Helsinki and was approved by the Ethical Committee of the first affiliated hospital. All participants or their relatives if they were unconscious had written informed consents before investigations were carried out.

Serum samples preparation

5 ml venous blood samples were collected from 54 septic patients and 41 SIRS patients within 12 hours of admission. With a first centrifugation (1,600 g, 10 min, 4°C) and a second centrifugation (16,000 g, 10 min, 4°C), Supernatant sera were separated and collected in 1.5 ml PCR tubes. Then, sera samples were stored in liquid nitrogen at -80°C until further qRT-PCR analysis.

RNA extraction and qRT-PCR

The microarray data was obtained from Gene Expression Omnibus (GEO; http://www.ncbi.nlm.nih.gov/geo/), and the GEO accession number is GSE47094. The expression of circulating miRNAs in mice with experimental sepsis

Table 3. The sequences of specific primers for qRT-PCR in this study

Gene	Primer sequences
Hsa-miR-195	
Forward	5'-CGTAGCAGCACAGAAAT-3'
Reverse	5'-GTGCAGGGTCCGAGGT-3'
Hsa-miR-300	
Forward	5'-TATACAAGGGCAGACTCTCTCT-3'
Reverse	5'-GTGCAGGTTCCGAGGT-3'
U6	
Forward	5'-CTCGCTTCGGCAGCACATATACT-3'
Reverse	5'-ACGCTTCACGAATTTGCGTGTC-3'

was analyzed using a miRNA array (The Mouse & Rat miRNA OneArray® v3). Three miRNAs, including miR-195, miR-300 and miR-468, were selected for further experimental validations.

According to the instruction, total RNA was isolated and extracted from 200 μl serum for each sample by QIAGEN miRNeasy Mini Kit (Qiagen, USA). Quantitative real-time reverse transcription polymerase chain reaction (qRT-PCR) was performed using ABI PRISM-7500 Sequence Detection System (Applied Biosystems, Foster City, CA, USA) with a 20 μl qRT-PCR system, including Forward Primer (0.6 μl), Reverse Primer (0.6 μl), cDNA (2 μl), ROX Reference Dye II (0.4 μl), SYBR Premix Ex Taq (10 μl), and ddH_2O (6.6 μl). The sequences of miR-195, miR-300 and miR-468 were obtained from https://www.ncbi.nlm.nih.gov/nuccore. The specific primers of miR-468 were purchased from Ribobio (Ribobio co., Ltd, China), and other primers for qRT-PCR in this study were listed in **Table 3**. The method of PCR cycling was 95°C for 5 min, followed by 40 cycles of 95°C for 15 s, 60°C for 30 s and 72°C for 30 s. The relative expression levels of three miRNAs were analyzed and calculated using $2^{-\Delta\Delta Ct}$ method ($Ct_{,target\ miRNA}$-$Ct_{,U6}$). In our study, U6 was applied as internal reference.

Survival analysis

A 28-day follow-up was conducted by hospitalized information or contacting with patients and their family members. Overall survival was defined as the ratio of patient alive and the total patient at the end of follow up. Survival curves were manufactured using the Kaplan-Meier method and differences in survival were evaluated by log-rank test.

Statistical analyses

Data were analyzed and performed using SPSS 19.0 software (SPSS Inc., Chicago, US) and presented by Graph PAD prism software 6.0 (GraphPad Software, Inc., US). Quantitative values are expressed as the means ± SD of each group of samples, and were analyzed by an unpaired Student's *t*-test or one-way ANOVA as appropriate. Qualitative data were analyzed by chi-square tests. Diagnostic accuracy was estimated using the receiver operating characteristic (ROC) procedure. According to the ROC curve, diagnosis cut-off points, specificity and sensitivity were analyzed and calculated. Spearman's rank-order correlation coefficient was applied for correlation analysis. Two-tailed P value <0.05 was regarded statistically significant.

Results

Serum level of miR-195 was up-regulated in sepsis

First, hierarchical clustering and heat map analysis showed that three miRNAs, including miR-195, miR-300, and miR-468, were differentially expressed in the whole blood samples of mice with experimental sepsis compared to healthy mice (**Figure 1A**). To validate the microarray analysis findings, 24 hours after admission to ICU, the serum expression of these three candidate miRNAs was detected through qRT-PCR in two cohorts of patients. We found that septic patients had a higher serum expression of miR-195 than that of control group (**Figure 1B**, P<0.001). In contrast, as shown in **Figure 1C** and **1D**, there were no significant differences in serum miR-300 and miR-468 expression between sepsis group and control group (all P>0.05). These results revealed that expression level of miR-195 in serum was elevated in sepsis.

Serum levels of miR-195 in septic patients with different infection sites

To investigate the differential expression of serum miR-195 with different infection sites, 54 septic patients were subsequently allocated into 5 subgroups according to their infection sites, including pulmonary group, abdominal group, urological group, brain-derived group and other group. However, as illustrated in **Figure 2**, no statistically significant difference

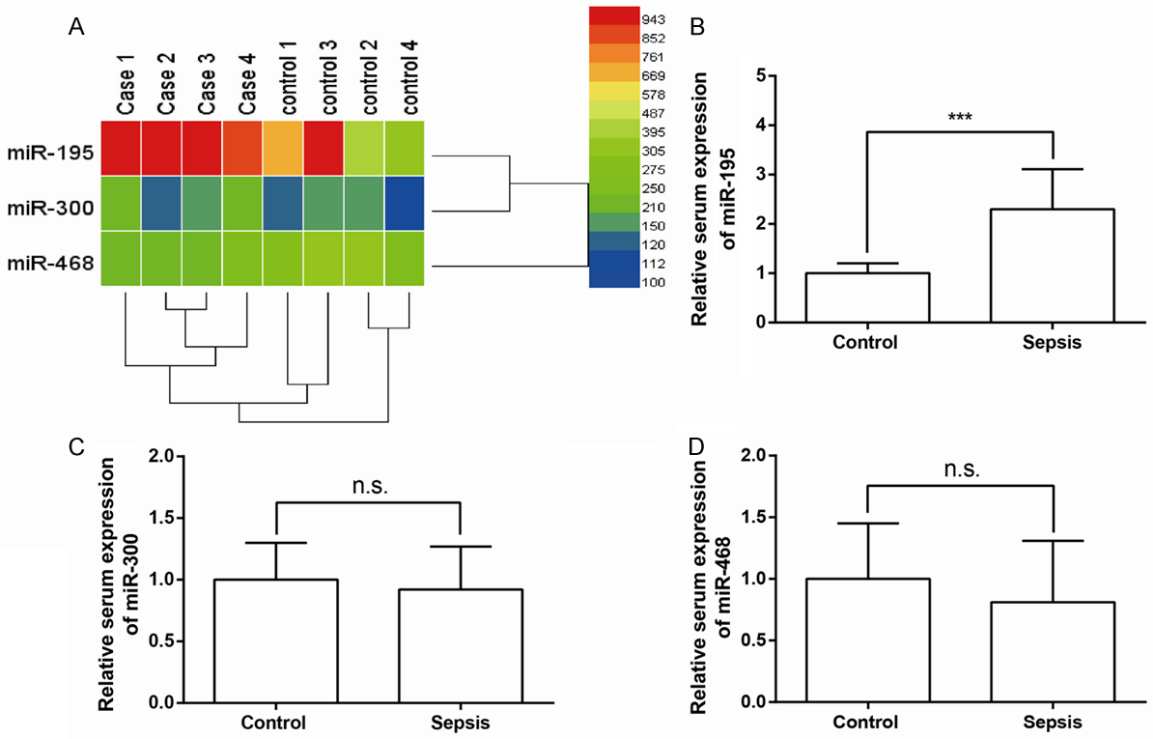

Figure 1. Expression levels of three miRNAs in serum. A. Hierarchical clustering and heat map analysis of three miRNAs that were differentially expressed in the whole blood samples of mice with experimental sepsis and healthy mice. The scale color from green (low expression) to red (high expression) indicates the expression levels of each miRNA. B. miR-195 expression levels in sera samples were analyzed. C. miR-300 expression levels in sera samples were analyzed. D. miR-468 expression levels in sera samples were analyzed. Data are expressed as the means ± SD of each group of samples. ***P<0.001; n. s., P>0.05.

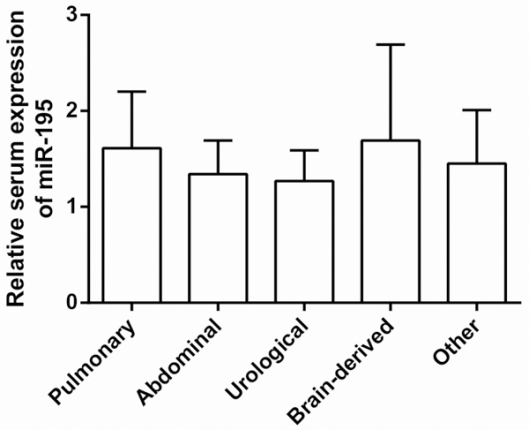

Figure 2. Serum levels of miR-195 in septic patients with different infection sites. Data are expressed as the means ± SD of each group of samples.

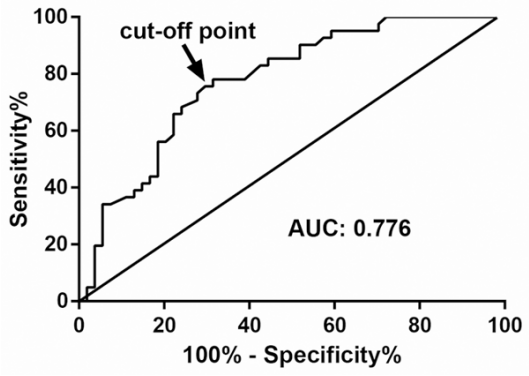

Figure 3. Receiver-operating characteristic (ROC) curve analysis using serum miR-195 for discriminating septic patients from patients with SIRS. X-axis refers to specificity and Y-axis refers to sensitivity.

in serum miR-195 expression was observed among these subgroups (P>0.05). Therefore, the results indicated that there was no significant association between serum levels of miR-195 and infection sites of septic patients.

Serum miR-195 as a biomarker in sepsis and its diagnostic value

With the result above, we aimed to determine whether serum miR-195 could be considered as a promising biomarker for sepsis diagnosis.

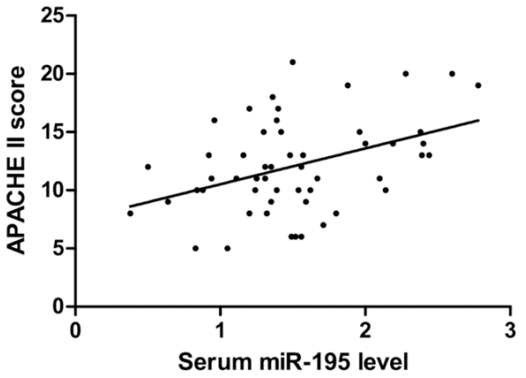

Figure 4. Correlation between serum miR-195 levels and APACHE II scores in septic patients. APACHE, Acute Physiology and Chronic Health Evaluation.

The levels of serum miR-195 in sepsis group and control group were calculated and analyzed through manufacturing ROC curve. As exhibited in **Figure 3**, the area under the ROC curve (AUC) of serum miR-195 was 0.776, with a 95% confidence interval (CI) of 0.6832-0.8687. We considered the best threshold was 1.245 through the Youden-index method, and specificity and sensitivity were 70.37% and 75.61%, respectively. Therefore, our data demonstrated that serum miR-195 might be a potential biomarker for the diagnosis of sepsis.

Additionally, Spearman's rank-order correlation coefficient analysis showed that the serum miR-195 levels were positively correlated with APACHE II scores of septic patients (**Figure 4**; r=0.417, *P*=0.002), indicating that serum miR-195 could represent the severity of septic condition.

The relationships between different levels of serum miR-195 and prognostic outcomes in septic patients

We next determined the correlation between serum miR-195 levels and prognosis of septic patients using the Kaplan-Meier method. 54 septic patients were allocated into low expression group (n=30) and high expression group (n=24) according to their serum miR-195 levels (**Figure 5A**). As demonstrated in **Figure 5B**, septic patients with high miR-195 level had relatively lower 28 d survival rates, compared with patients with low miR-195 level, but the difference was not statistically significant (*P*=0.113). The above data indicated that the miR-195 level was failed to predict the prognosis of septic patients.

Discussion

RNA extraction is one of the most important methods in molecular biology, which was extensively used in multiple scientific researches. Actually, once isolated from the tissues, cells or other samples, RNAs are susceptible to degradation [20]. Therefore, RNase-free technique serves a crucial role in inhibiting endogenous RNases function in RNA extraction. MiRNAs is a class of small noncoding RNAs. Lee *et al.* [21] originally found that lin-4 in C. elegans could repress the expression level of lin-14 protein via binding with the 3'UTR of lin-14 mRNA. Then, increasing evidence demonstrated that miRNAs were associated with a number of human diseases, such as tumor [22], respiratory viral infection [23], Parkinson's disease [24], congenital heart disease [25] and alcoholic hepatitis [26]. Besides, miRNAs could be easily isolated from blood, serum and other body fluids. Moreover, because of resistance to environmental changes, miRNAs are highly stable in body fluids, such as plasma and serum [27]. Unlike genomic DNA, which is static, RNA expression can dynamically change over healthy and diseased states and, thus, can provide real-time information concerning cellular function. Thus, miRNAs in plasma or serum were characterized as potential biomarkers for diagnosis of various human diseases [28-30].

The emerging study revealed that a series of miRNAs participated in the pathogenesis and progression of sepsis. For example, Wang *et al.* [31] detected and analyzed the serum expression of miR-297 and miR-574-5p and found that serum miR-574-5p was significantly correlated to unfavorable outcomes in septic patients and was a promising prognostic biomarker for sepsis. In the present study, we found that serum miR-195 was obviously up-regulated in sepsis group in comparison with that in control group and could be a potential diagnostic biomarker of sepsis. Similarly, Zheng and coworkers [32] established the septic mice models through injecting feces into the abdominal cavity of mice and found that miR-195 expression was increased in the lung and liver of septic mice and up-regulation of miR-195 could promote cell apoptosis *in vitro*. In addi-

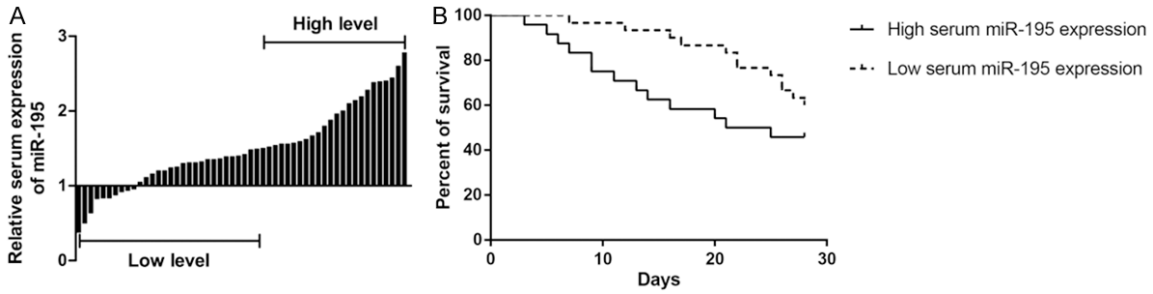

Figure 5. Prognostic significance of serum miR-195 level in septic patients. A. The fold changes of relative serum miR-195 expression of each septic patient. B. Kaplan-Meier survival curves of septic patients based on serum miRNA-195 expression level.

tion, Wu *et al.* [33] revealed that significantly up-regulated miR-195 expression in the whole blood was found in septic mice compared with that in sham-operated mice through microarray analysis, which might be correlated to the sepsis-related pathophysiological responses, including inflammation, shock and ileus. NF-κB, a critical regulatory factor of genes associated to inflammation, serves a crucial role in the development and progression of sepsis via promoting the transcription of inflammatory cytokines and modulating the inflammatory cascade reactions [34]. Previous research has reported also that miR-195 might be involved in the NF-κB signaling pathway by way of the direct targeting of IKKα and TAB3 [35].

As we all known, the prognosis of sepsis was still dissatisfied. The extensive clinical applications of biomarkers might serve a critical role in the early diagnosis and evaluation of disease condition of sepsis. A wide range of biological moleculars, including immature platelet fraction [36], blood lactic acid [37], C-reactive protein (CRP) [38] and procalcitonin (PCT) [39], were associated with the prognostic outcomes of septic patients. Moreover, the recent studies revealed that some miRNAs also were significantly correlated to the prognosis of sepsis [40-42]. Zheng *et al.* [32] further investigation demonstrated that multiple-organ injury and cell apoptosis were obviously reduced and the survival rate was increased after silencing of miR-195 in mouse models of sepsis. In our study, the results revealed that the patients with high levels of serum miR-195 had a lower survival rate than those with low level of serum miR-195. However, it is intriguing to find that the difference was not statistically significant. We considered that this difference might be caused due to species differences between mice and human beings.

Of note, the present study has some limitations. Firstly, our study had included insufficient number of septic patients and SIRS patients from a single center, which might impair credibility of data in the current study. Secondly, selection bias was inevitable during the inclusion of septic patients and SIRS patients. Further investigations are required to evaluate the serum miR-195 levels in a larger scale of population and to explore the underlying mechanisms how miR-195 plays a role in sepsis and the interaction between miR-195 and downstream genes, including NF-κB signaling pathway, in experimental animal models.

To conclude, the results of our study are the first to show that expression of miR-195 in serum was remarkably increased in septic patients than that in SIRS patients. Despite the weak prognostic value, serum miR-195 could serve as a potential and useful noninvasive biomarker for the patients with sepsis.

Disclosure of conflict of interest

None.

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References

[1] Fink MP and Warren HS. Strategies to improve drug development for sepsis. Nat Rev Drug Discov 2014; 13: 741-758.

[2] Tonolini M and Ippolito S. Cross-sectional imaging of complicated urinary infections affecting the lower tract and male genital organs. Insights Imaging 2016; 7: 689-711.
[3] Deng S, Zhu H, Wang K and Cao T. Procalcitonin as a marker of sepsis and outcome in patients with neurotrauma: an observation study. BMC Anesthesiol 2013; 13: 48.
[4] Burt RK, Oliveira MC, Shah SJ, Moraes DA, Simoes B, Gheorghiade M, Schroeder J, Ruderman E, Farge D, Chai ZJ, Marjanovic Z, Jain S, Morgan A, Milanetti F, Han X, Jovanovic B, Helenowski IB and Voltarelli J. Cardiac involvement and treatment-related mortality after non-myeloablative haemopoietic stem-cell transplantation with unselected autologous peripheral blood for patients with systemic sclerosis: a retrospective analysis. Lancet 2013; 381: 1116-1124.
[5] Schrier RW and Wang W. Acute renal failure and sepsis. N Engl J Med 2004; 351: 159-169.
[6] Gaieski DF, Edwards JM, Kallan MJ and Carr BG. Benchmarking the incidence and mortality of severe sepsis in the united states. Crit Care Med 2013; 41: 1167-1174.
[7] Tang BM, Eslick GD, Craig JC and McLean AS. Accuracy of procalcitonin for sepsis diagnosis in critically ill patients: systematic review and meta-analysis. Lancet Infect Dis 2007; 7: 210-217.
[8] Clerico A and Plebani M. Biomarkers for sepsis: an unfinished journey. Clin Chem Lab Med 2013; 51: 1135-1138.
[9] Pinweha P, Rattanapornsompong K, Charoensawan V and Jitrapakdee S. MicroRNAs and oncogenic transcriptional regulatory networks controlling metabolic reprogramming in cancers. Comput Struct Biotechnol J 2016; 14: 223-233.
[10] Issler O and Chen A. Determining the role of microRNAs in psychiatric disorders. Nat Rev Neurosci 2015; 16: 201-212.
[11] Mehta A and Baltimore D. MicroRNAs as regulatory elements in immune system logic. Nat Rev Immunol 2016; 16: 279-294.
[12] Ward PA. The bipolar role of miR-466l in inflammation. Immunity 2013; 39: 801-802.
[13] Sun X, Icli B, Wara AK, Belkin N, He S, Kobzik L, Hunninghake GM, Vera MP, Blackwell TS, Baron RM and Feinberg MW. MicroRNA-181b regulates NF-kappaB-mediated vascular inflammation. J Clin Invest 2012; 122: 1973-1990.
[14] Han Y, Dai QC, Shen HL and Zhang XW. Diagnostic value of elevated serum miRNA-143 levels in sepsis. J Int Med Res 2016; 44: 875-81.
[15] Liu S, Liu C, Wang Z, Huang J and Zeng Q. microRNA-23a-5p acts as a potential biomarker for sepsis-induced acute respiratory distress syndrome in early stage. Cell Mol Biol (Noisy-legrand) 2016; 62: 31-37.
[16] Tacke F, Roderburg C, Benz F, Cardenas DV, Luedde M, Hippe HJ, Frey N, Vucur M, Gautheron J, Koch A, Trautwein C and Luedde T. Levels of circulating miR-133a are elevated in sepsis and predict mortality in critically ill patients. Crit Care Med 2014; 42: 1096-1104.
[17] Min M, Peng LH, Sun G, Guo MZ, Qiu ZW and Yang YS. Aquaporin 8 expression is reduced and regulated by microRNAs in patients with ulcerative colitis. Chin Med J (Engl) 2013; 126: 1532-1537.
[18] Levy MM, Fink MP, Marshall JC, Abraham E, Angus D, Cook D, Cohen J, Opal SM, Vincent JL, Ramsay G; SCCM/ESICM/ACCP/ATS/SIS. 2001 SCCM/ESICM/ACCP/ATS/SIS international sepsis definitions conference. Crit Care Med 2003; 31: 1250-1256.
[19] Knaus WA, Draper EA, Wagner DP and Zimmerman JE. APACHE II: a severity of disease classification system. Crit Care Med 1985; 13: 818-829.
[20] Tan SC and Yiap BC. DNA, RNA, and protein extraction: the past and the present. J Biomed Biotechnol 2009; 2009: 574398.
[21] Lee RC, Feinbaum RL and Ambros V. The C. elegans heterochronic gene lin-4 encodes small RNAs with antisense complementarity to lin-14. Cell 1993; 75: 843-854.
[22] Yin G, Zhou H, Xue Y, Yao B and Zhao W. MicroRNA-340 promotes the tumor growth of human gastric cancer by inhibiting cyclin G2. Oncol Rep 2016; 36: 1111-8.
[23] Tahamtan A, Inchley CS, Marzban M, Tavakoli-Yaraki M, Teymoori-Rad M, Nakstad B and Salimi V. The role of microRNAs in respiratory viral infection: friend or foe? Rev Med Virol 2016; 26: 389-407.
[24] Nair VD and Ge Y. Alterations of miRNAs reveal a dysregulated molecular regulatory network in parkinson's disease striatum. Neurosci Lett 2016; 629: 99-104.
[25] Li FF, Deng X, Zhou J, Yan P, Zhao EY and Liu SL. Characterization of human bone morphogenetic protein gene variants for possible roles in congenital heart disease. Mol Med Rep 2016; 14: 1459-64.
[26] Liu Y, Chen SH and Li YM. [Differential expression profiles of genes and miRNAs in alcoholic hepatitis]. Zhonghua Gan Zang Bing Za Zhi 2012; 20: 883-887.
[27] Ortiz-Quintero B. Cell-free microRNAs in blood and other body fluids, as cancer biomarkers. Cell Prolif 2016; 49: 281-303.
[28] Kinoshita T, Yip KW, Spence T and Liu FF. MicroRNAs in extracellular vesicles: potential cancer biomarkers. J Hum Genet 2017; 62: 67-74.
[29] Navickas R, Gal D, Laucevicius A, Taparauskaite A, Zdanyte M and Holvoet P. Identifying circulating microRNAs as biomarkers of cardio-

vascular disease: a systematic review. Cardiovasc Res 2016; 111: 322-37.
[30] Kumar S and Reddy PH. Are circulating microRNAs peripheral biomarkers for alzheimer's disease? Biochim Biophys Acta 2016; 1862: 1617-1627.
[31] Wang H, Meng K, Chen W, Feng D, Jia Y and Xie L. Serum miR-574-5p: a prognostic predictor of sepsis patients. Shock 2012; 37: 263-267.
[32] Zheng D, Yu Y, Li M, Wang G, Chen R, Fan GC, Martin C, Xiong S and Peng T. Inhibition of microRNA 195 prevents apoptosis and multiple-organ injury in mouse models of sepsis. J Infect Dis 2016; 213: 1661-1670.
[33] Wu SC, Yang JC, Rau CS, Chen YC, Lu TH, Lin MW, Tzeng SL, Wu YC, Wu CJ and Hsieh CH. Profiling circulating microRNA expression in experimental sepsis using cecal ligation and puncture. PLoS One 2013; 8: e77936.
[34] Adamzik M, Schafer S, Frey UH, Becker A, Kreuzer M, Winning S, Frede S, Steinmann J, Fandrey J, Zacharowski K, Siffert W, Peters J and Hartmann M. The NFKB1 promoter polymorphism (-94ins/delATTG) alters nuclear translocation of NF-kappaB1 in monocytes after lipopolysaccharide stimulation and is associated with increased mortality in sepsis. Anesthesiology 2013; 118: 123-133.
[35] Ding J, Huang S, Wang Y, Tian Q, Zha R, Shi H, Wang Q, Ge C, Chen T, Zhao Y, Liang L, Li J and He X. Genome-wide screening reveals that miR-195 targets the TNF-alpha/NF-kappaB pathway by down-regulating IkappaB kinase alpha and TAB3 in hepatocellular carcinoma. Hepatology 2013; 58: 654-666.
[36] Muronoi T, Koyama K, Nunomiya S, Lefor AK, Wada M, Koinuma T, Shima J and Suzukawa M. Immature platelet fraction predicts coagulopathy-related platelet consumption and mortality in patients with sepsis. Thromb Res 2016; 144: 169-175.
[37] Yan HP, Lu XL, Qiu J, Liu PP, Zuo C and Zhu YM. [Value of blood lactic acid in evaluating disease severity and prognosis in children with sepsis]. Zhongguo Dang Dai Er Ke Za Zhi 2016; 18: 506-510.
[38] Ryu JA, Yang JH, Lee D, Park CM, Suh GY, Jeon K, Cho J, Baek SY, Carriere KC and Chung CR. Clinical usefulness of procalcitonin and c-reactive protein as outcome predictors in critically ill patients with severe sepsis and septic shock. PLoS One 2015; 10: e0138150.
[39] Huang MY, Chen CY, Chien JH, Wu KH, Chang YJ and Wu HP. Serum procalcitonin and procalcitonin clearance as a prognostic biomarker in patients with severe sepsis and septic shock. Biomed Res Int 2016; 2016: 1758501.
[40] Benz F, Roy S, Trautwein C, Roderburg C and Luedde T. Circulating MicroRNAs as biomarkers for sepsis. Int J Mol Sci 2016; 17.
[41] Wang HJ, Zhang PJ, Chen WJ, Jie D, Dan F, Jia YH and Xie LX. Characterization and identification of novel serum microRNAs in sepsis patients with different outcomes. Shock 2013; 39: 480-487.
[42] Roderburg C, Luedde M, Vargas Cardenas D, Vucur M, Scholten D, Frey N, Koch A, Trautwein C, Tacke F and Luedde T. Circulating microRNA-150 serum levels predict survival in patients with critical illness and sepsis. PLoS One 2013; 8: e54612.